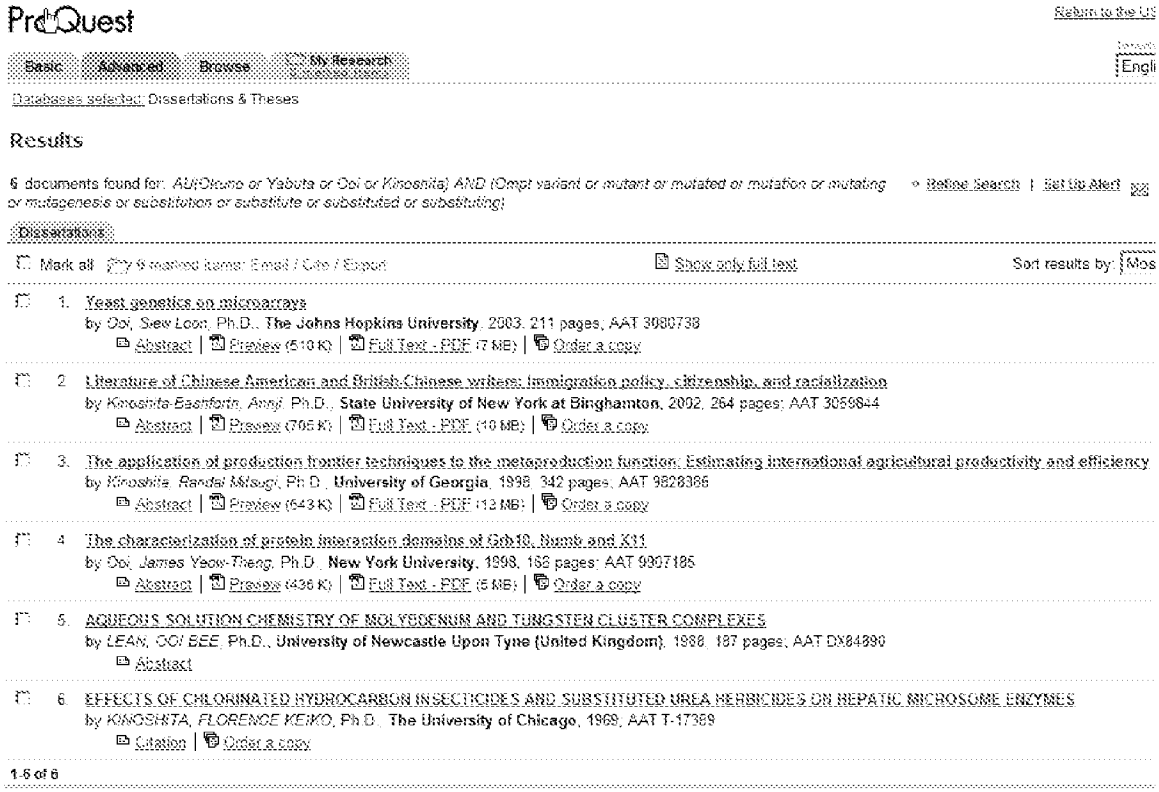
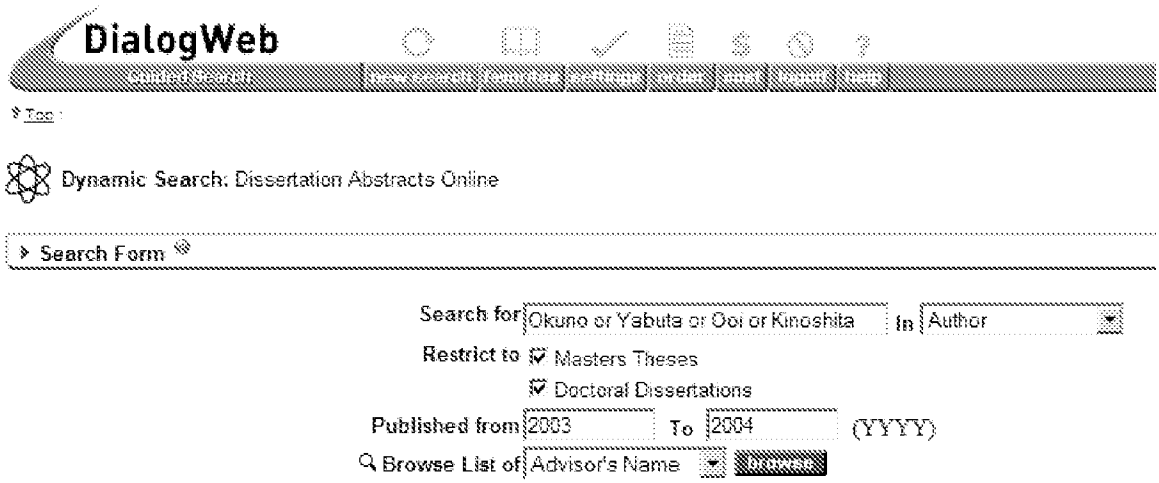
I searched the following databases using author and keyword suggested by the examiner:





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We found 1 article by title matching your search:

Appl Environ Microbiol. 2004 Jan;70(1):76-86.

Utilization of Escherichia coli outer-membrane endoprotease OmpT variants as processing enzymes for production of peptides from designer fusion proteins.

Ohnuno K, Yabuta M, Doi T, Kinoshita S.

Institute for Medicinal Research and Development, Daiichi Suntory Pharma Co., Ltd., Akaiwa, Chiyoda-machi, Ohra-gun, Gunma 370-0503, Japan. Kazuaki_Ohnuno@dsup.co.jp

&Abstract&

Escherichia coli outer-membrane endoprotease OmpT has suitable properties for processing fusion proteins to produce peptides and proteins. However, utilization of this protease for such production has been restricted due to its generally low cleavage efficiency at Arg (or Lys)-Xaa, where Xaa is a nonbasic N-terminal amino acid of a target polypeptide. The objective of this study was to generate a specific and efficient OmpT protease and to utilize it as a processing enzyme for producing various peptides and proteins by converting its substrate specificity. Since OmpT Asp(97) is proposed to interact with the P1' amino acid of its substrates, OmpT variants with variations at Asp(97) were constructed by replacing this amino acid with 19 natural amino acids to alter the cleavage specificity at Arg (P1)-Xaa (P1'). The variant OmpT that had a methionine at this position, but not the wild-type OmpT, efficiently cleaved a fusion protein containing the amino acid sequence -Arg-Arg-Arg-Ala-Arg downward arrow motilin, in which motilin is a model peptide with a phenylalanine at the N terminus. The OmpT variants with leucine and histidine at position 97 were useful in releasing human adrenocorticotropic hormone (1-24) (serine at the N terminus) and human calcitonin precursor (cysteine at the N terminus), respectively, from fusion proteins. Motilin was produced by this method and was purified up to 99.0% by two chromatographic steps; the yield was 160 mg/liter of culture. Our novel method in which the OmpT variants are used could be employed for production of various peptides and proteins.

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STAT- MEDLINE
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IP - 1
DP - 2004 Jan
TI - Utilization of Escherichia coli outer-membrane endoprotease OmpT variants as
processing enzymes for production of peptides from designer fusion proteins.
PG - 76-86
AB - Escherichia coli outer-membrane endoprotease OmpT has suitable properties for
processing fusion proteins to produce peptides and proteins. However, utilization
of this protease for such production has been restricted due to its generally low
cleavage efficiency at Arg (or Lys)-Xaa, where Xaa is a nonbasic N-terminal amino
acid of a target polypeptide. The objective of this study was to generate a
specific and efficient OmpT protease and to utilize it as a processing enzyme for
producing various peptides and proteins by converting its substrate specificity.

Since OmpT Asp(97) is proposed to interact with the P1' amino acid of its substrates, OmpT variants with variations at Asp(97) were constructed by replacing this amino acid with 19 natural amino acids to alter the cleavage specificity at Arg (P1)-Xaa (P1'). The variant OmpT that had a methionine at this position, but not the wild-type OmpT, efficiently cleaved a fusion protein containing the amino acid sequence -Arg-Arg-Arg-Ala-Arg downward arrow motilin, in which motilin is a model peptide with a phenylalanine at the N terminus. The OmpT variants with leucine and histidine at position 97 were useful in releasing human adrenocorticotropic hormone (1-24) (serine at the N terminus) and human calcitonin precursor (cysteine at the N terminus), respectively, from fusion proteins. Motilin was produced by this method and was purified up to 99.0% by two chromatographic steps; the yield was 160 mg/liter of culture. Our novel method in which the OmpT variants are used could be employed for production of various peptides and proteins.

AD - Institute for Medicinal Research and Development, Daiichi Suntory Pharma Co., Ltd., Akaiwa, Chiyoda-machi, Ohra-gun, Gunma 370-0503, Japan. Kazuaki_Okuno@dsup.co.jp
FAU - Okuno, Kazuaki
AU - Okuno K
FAU - Yabuta, Masayuki
AU - Yabuta M
FAU - Ooi, Toshihiko
AU - Ooi T
FAU - Kinoshita, Shinichi
AU - Kinoshita S

LA - eng
PT - Journal Article
PL - United States
TA - Appl Environ Microbiol
JT - Applied and environmental microbiology
JID - 7605801
RN - 0 (Bacterial Outer Membrane Proteins)
RN - 0 (Escherichia coli Proteins)
RN - 0 (Peptides)
RN - 0 (Porins)
RN - 0 (Recombinant Fusion Proteins)
RN - 0 (ompT protein, E coli)
RN - 52906-92-0 (Motilin)
RN - EC 3.4.- (Peptide Hydrolases)
RN - EC 3.4.21.- (Serine Endopeptidases)
SB - IM
MH - Amino Acid Sequence
MH - Bacterial Outer Membrane Proteins

MH - Escherichia coli/*enzymology/genetics
MH - Escherichia coli Proteins
MH - *Genetic Variation
MH - Humans
MH - Inclusion Bodies/metabolism
MH - Molecular Sequence Data
MH - Motilin/genetics/metabolism
MH - Mutation
MH - Peptide Hydrolases
MH - Peptides/*metabolism
MH - Porins/genetics/*metabolism
MH - Recombinant Fusion Proteins
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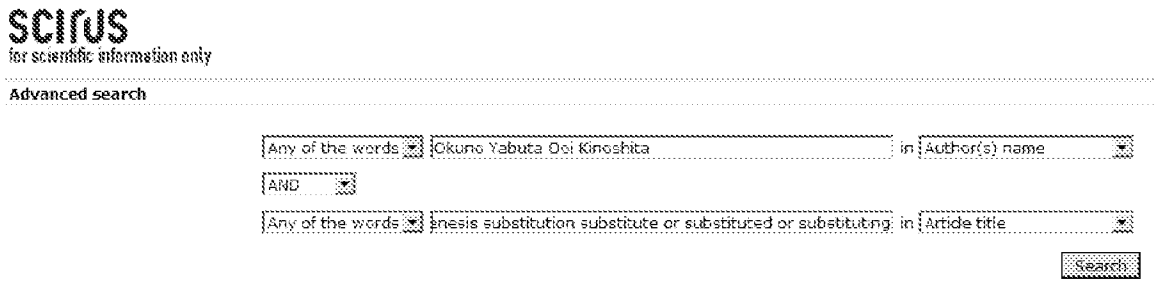
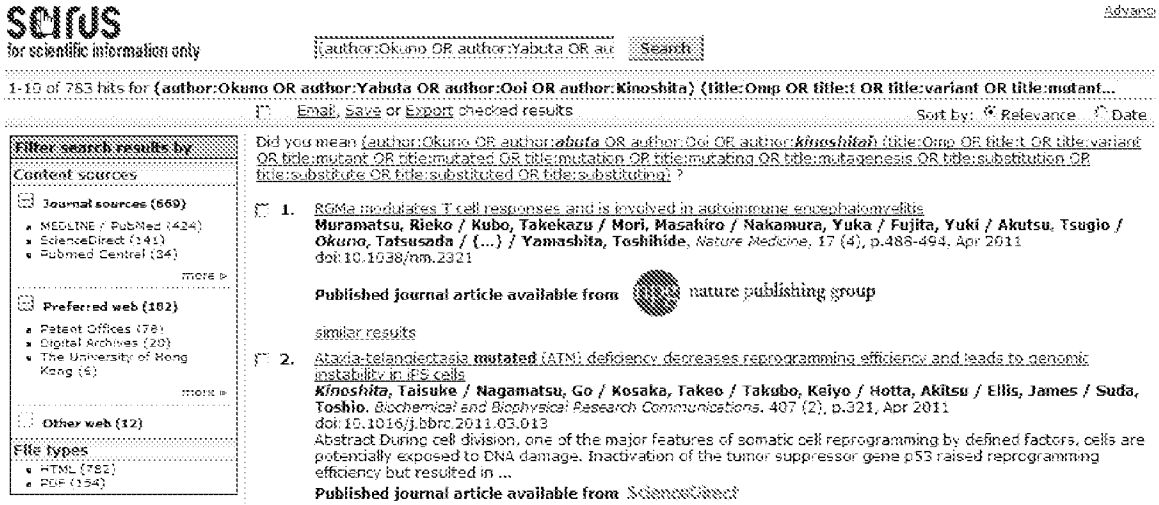
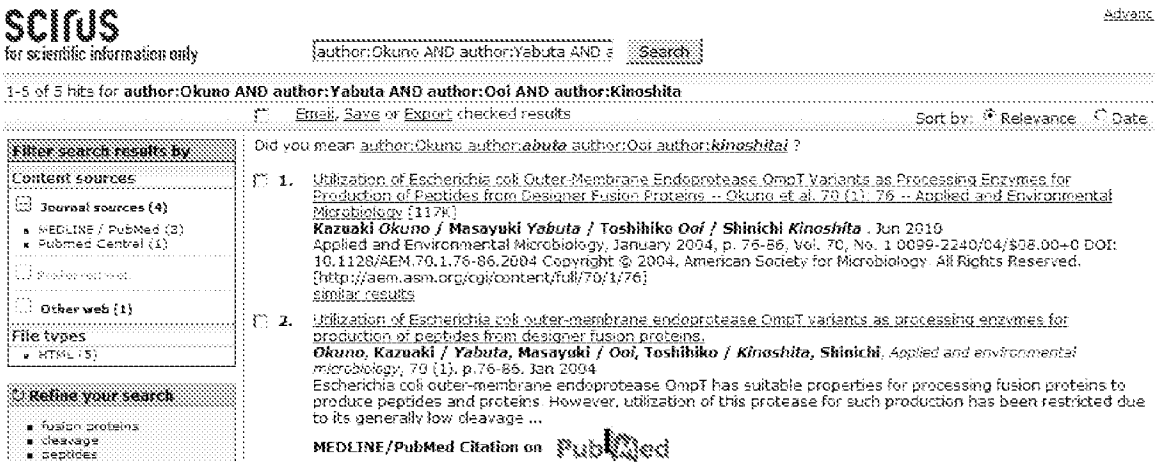
PST - ppublish
SO - Appl Environ Microbiol. 2004 Jan;70(1):76-86.





Same database using AND for authors and no words in the title section:



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From the Scirus second link above: